

## AMENDMENTS TO THE SPECIFICATION

On page 1, between lines 2 and 3, please insert the following:

### Cross-Reference to Related Applications

The present application is a divisional application of U.S. serial no. 09/636,060 filed August 10, 2000, allowed, which claims the benefit of U.S. provisional application serial number 60/153,831 filed September 14, 1999.

Page 14, line 14 to page 15, line 14, please replace the paragraph with the following paragraph:

To obtain isolated and purified GlcNAc-phosphotransferase and its subunits and the nucleic acid molecules encoding the enzyme according to the present invention, bovine GlcNAc phosphotransferase was obtained and analyzed as follows. Splenocytes from mice immunized with a partially purified preparation of bovine GlcNAc-phosphotransferase were fused with myeloma cells to generate a panel of hybridomas. Hybridomas secreting monoclonal antibodies specific for GlcNAc-phosphotransferase were identified by immunocapture assay. In this assay, antibodies which could capture GlcNAc-phosphotransferase from a crude source were identified by assay of immunoprecipitates with a specific GlcNAc-phosphotransferase enzymatic assay. Hybridomas were subcloned twice, antibody produced in ascites culture, coupled to a solid support and evaluated for immunoaffinity chromatography. Monoclonal PT18-Emphaze was found to allow a single step purification of GlcNAc-phosphotransferase to homogeneity. Bao, *et.al.*, The Journal of Biological Chemistry, Vol. 271, Number 49, Issue of December 6, 1996, pp. 31437-31445 relates to a method for the purification of bovine UDP-N-acetylglucosamine:Lysosomal-enzyme N-Acetylglucosamine-1-phosphotransferase and proposes a hypothetical subunit structure for the protein. Bao, *et. al.*, *The Journal of Biological Chemistry*, Vol. 271,

Number 49, Issue of December 6, 1996, pp. 31446-31451. Using this technique, the enzyme was purified 488,000-fold in 29% yield. The eluted GlcNAc-phosphotransferase has a specific activity of  $>10^6$ , preferably  $>5 \times 10^6$ , more preferably  $>12 \times 10^6$  pmol/h/mg and is apparently a homogenous, multi-subunit enzyme based on silver-stained SDS-PAGE. The monoclonal antibody labeled PT18 was selected for use in further experiments. A hybridoma secreting monoclonal antibody PT 18 was deposited with the American Type Culture Collection, 10801 University Blvd., Manassas, VA 20110 on August 29, 2000 and assigned ATCC Accession No. [[\_\_\_\_\_]]PTA 2432.

Page 20, line 23 to page 21, line 22, please replace the paragraph with the following paragraph:

Following a second intravenous boost with phosphodiester  $\alpha$ -GlcNAcase, the spleen was removed and splenocytes fused with SP2/0 myeloma cells according to our modifications (Bag, M., Booth J. L., *et al.* (1996). "Bovine UDP-N-acetylglucosamine: lysosomal enzyme N-acetylglucosamine-1-phosphotransferase. I. Purification and subunit structure." *Journal of Biological Chemistry* 271: 31437 - 31445) of standard techniques; Harlow, E. and Lane, D. (1988). *Antibodies: a laboratory manual*, Cold Spring Harbor Laboratory). The fusion was plated in eight 96-well plates in media supplemented with recombinant human IL-6 (Bazin, R. and Lemieux, R. (1989). "Increased proportion of B cell hybridomas secreting monoclonal antibodies of desired specificity in cultures containing macrophage-derived hybridoma growth factor (IL-6)." *Journal of Immunological Methods* 116: 245 - 249) and grown until hybridomas were just visible. Forty-eight pools of 16-wells were constructed and assayed for antiphosphodiester  $\alpha$ -GlcNAcase activity using the capture assay. Four pools were positive. Subpools of 4-wells were then constructed from the wells present in the positive 16-well pools. Three of the four 16-well pools contained a single 4-well pool with anti-phosphodiester  $\alpha$ -GlcNAcase activity. The 4 single wells making up the 4-well pools

were then assayed individually identifying the well containing the anti-phosphodiester  $\alpha$ -GlcNAcase secreting hybridomas. Using the capture assay, each hybridoma was subcloned twice and antibody prepared by ascites culture. Monoclonals UC2 and UC3 were found to be low affinity antibodies. UC1, a high affinity IgG monoclonal antibody, was prepared by ascites culture and immobilized on Emphaze for purification of phosphodiester  $\alpha$ -GlcNAcase. The monoclonal antibody labeled UC1 was selected for use in further experiments. A hybridoma secreting monoclonal antibody UC1 was deposited with the American Type Culture Collection, 10801 Univerisity Blvd., Manassas, VA 20110 on August 29, 2000 and assigned ATCC Accession No. [[\_\_\_\_\_]] PTA 2431.

Page 76 (Abstract), after the last line, beginning on a new page, please insert the attached Sequence Listing.